

SPECTRAL IMAGING APPARATUS AND METHODOLOGY

## ABSTRACT

5 A method and apparatus for an improved spectral imaging system is provided. The system is capable of measuring the fluorescence, luminescence, or absorption at selected locations on a sample plate. The emissions detection subassembly can tune to any wavelength within a continuum of wavelengths utilizing an interferometric spectral discriminator. The interferometric spectral discriminator creates an interferogram from which the wavelength spectra for each pixel of the array  
10 can be calculated, typically using Fourier transform analysis. In one aspect, the chromatic accuracy of the system is calibrated using a calibration slit placed in the input aperture of the input relay lens but outside of the sample image. The slit is illuminated using a source of known wavelength. The fringe count versus the wavelength of the slit illumination source is monitored and used to calibrate the  
15 spectral discriminator. In another aspect, a transparent optic is included in the interferometric spectral discriminator that can be inserted into the beam path whenever a monochrome image of the sample is required. The optic produces a large offset in the legs of the interferometer resulting in the fringe density becoming too large to resolve by the individual pixels of the detector array. In another aspect, the interferometric  
20 spectral discriminator includes a polarizing beam splitter. The polarizing beam splitter preferentially reflects one polarization while preferentially transmitting a second polarization, thus achieving improved efficiency while minimizing ghosting. In another aspect, a metaphase finder is used to locate areas of interest. The sample plate containing the material of interest is illuminated with light of a wavelength determined  
25 to preferentially scatter from objects the size of the metaphase spreads. The intensity of the scattered light versus the location on the sample plate is monitored and used to locate the areas of interest. Preferably the sample plate is also illuminated by light of a second wavelength which is not preferentially scattered by the objects of interest, thus representing the background scatter. By subtracting the background scatter from the  
30 primary scattered light, improved object discrimination is achieved.